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N2O and NO dynamics in AOB-enriched and mixed-culture biomass: experimental observations and model calibration

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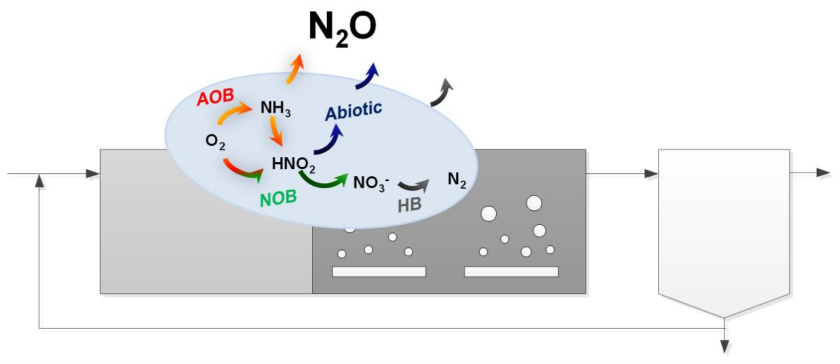
Introduction

- Nitrous oxide (N_2O) emissions during nitrogen removal in wastewater treatment operations can compromise the environmental impact of new energy-saving technologies.
- Current process modelling efforts aim to reproduce N_2O experimental data with mathematical equations, structuring our understanding of the system.
- A mathematical model structure that describes N_2O production during biological nitrogen removal[1] is calibrated for two biomasses representative of wastewater treatment operations: AOB-enriched and mixed culture.
- Extant respirometric assays are used to monitor N_2O, NO and DO dynamics.

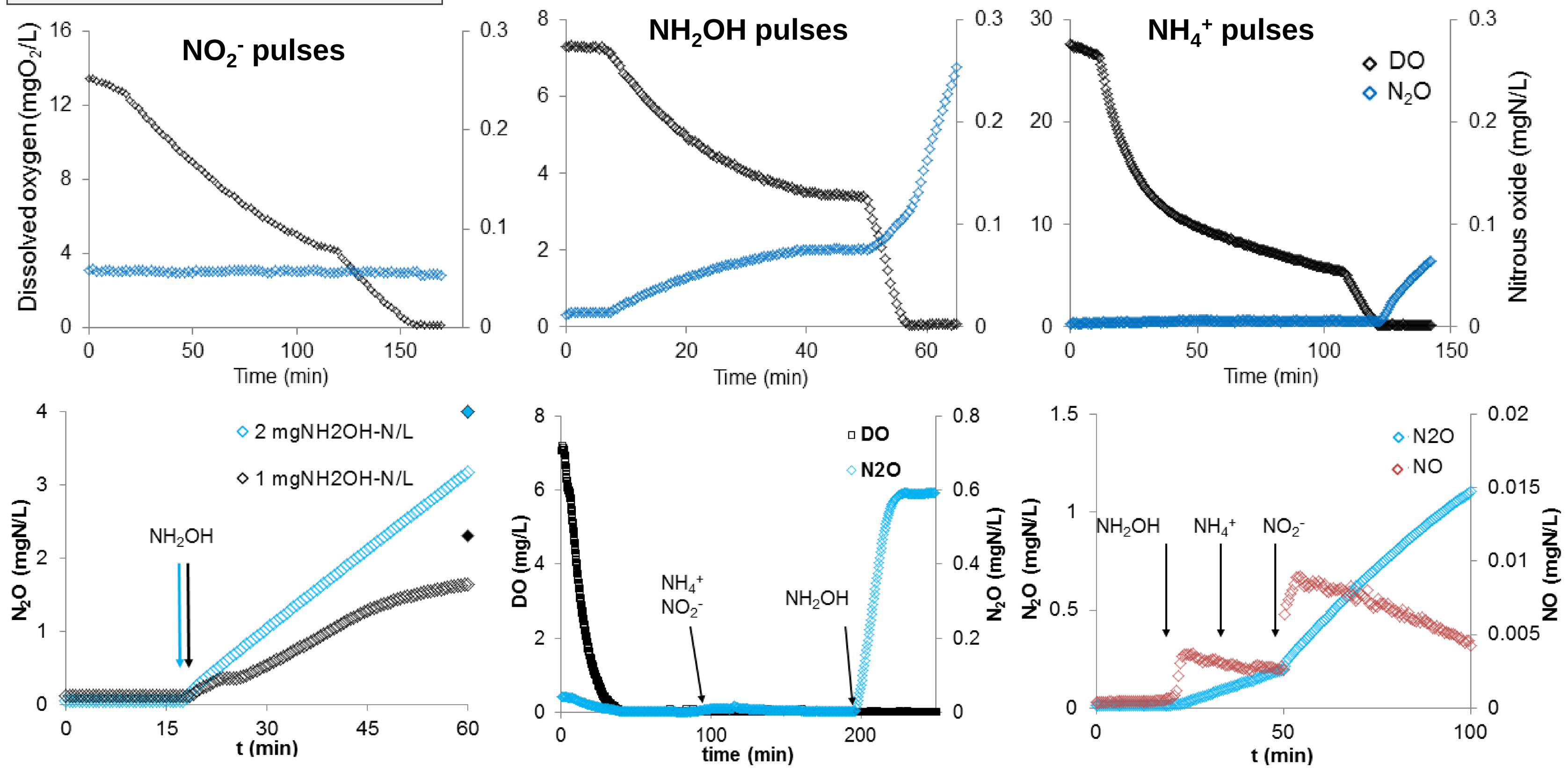


Obj_1 → Quantify N_2O dynamics via extant respirometric assays from two biomasses: AOB-enriched and mixed liquor.

Obj_2 → Calibrate the NDHA model[1] to describe N-removing processes and N_2O production for wastewater treatment operations.

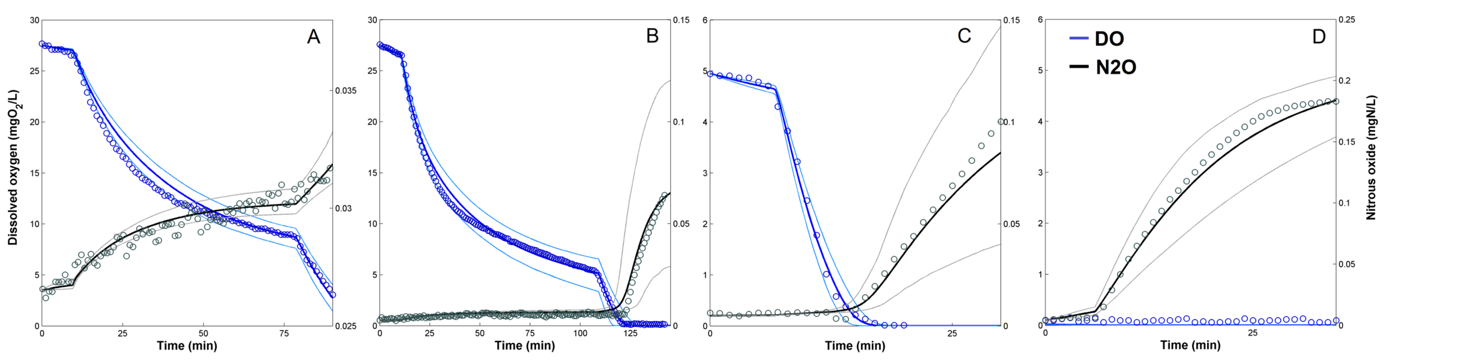
Experimental and Modelling results

AOB-enriched



Top – Aerobic experiments. Dissolved oxygen and N_2O concentrations during NO_2^- (left), NH_2OH (middle) and NH_4^+ (right) pulses (1-4 mgN/L).

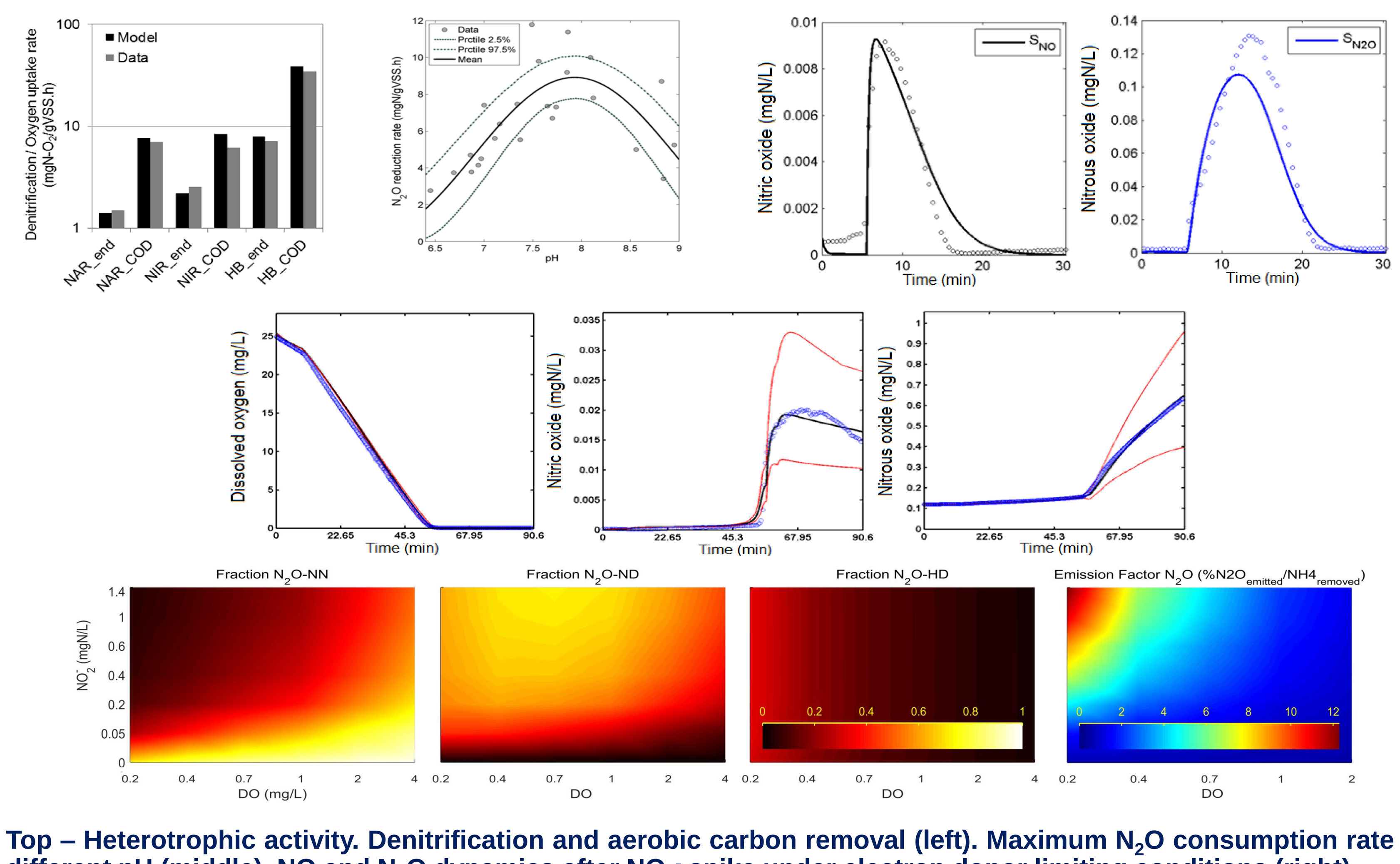
Bottom – Anoxic experiments. NH_2OH oxidation at excess NO_2^- (left), effect of NO_2^- and NH_4^+ on N_2O production (middle), effect of NH_2OH and NO_2^- oxidation on N_2O and NO production (right).



Experimental and modelling results for DO and N_2O during NH_4^+ oxidation at high DO (A), from high to low DO (B, C) and anoxic NH_2OH oxidation (D).[2]

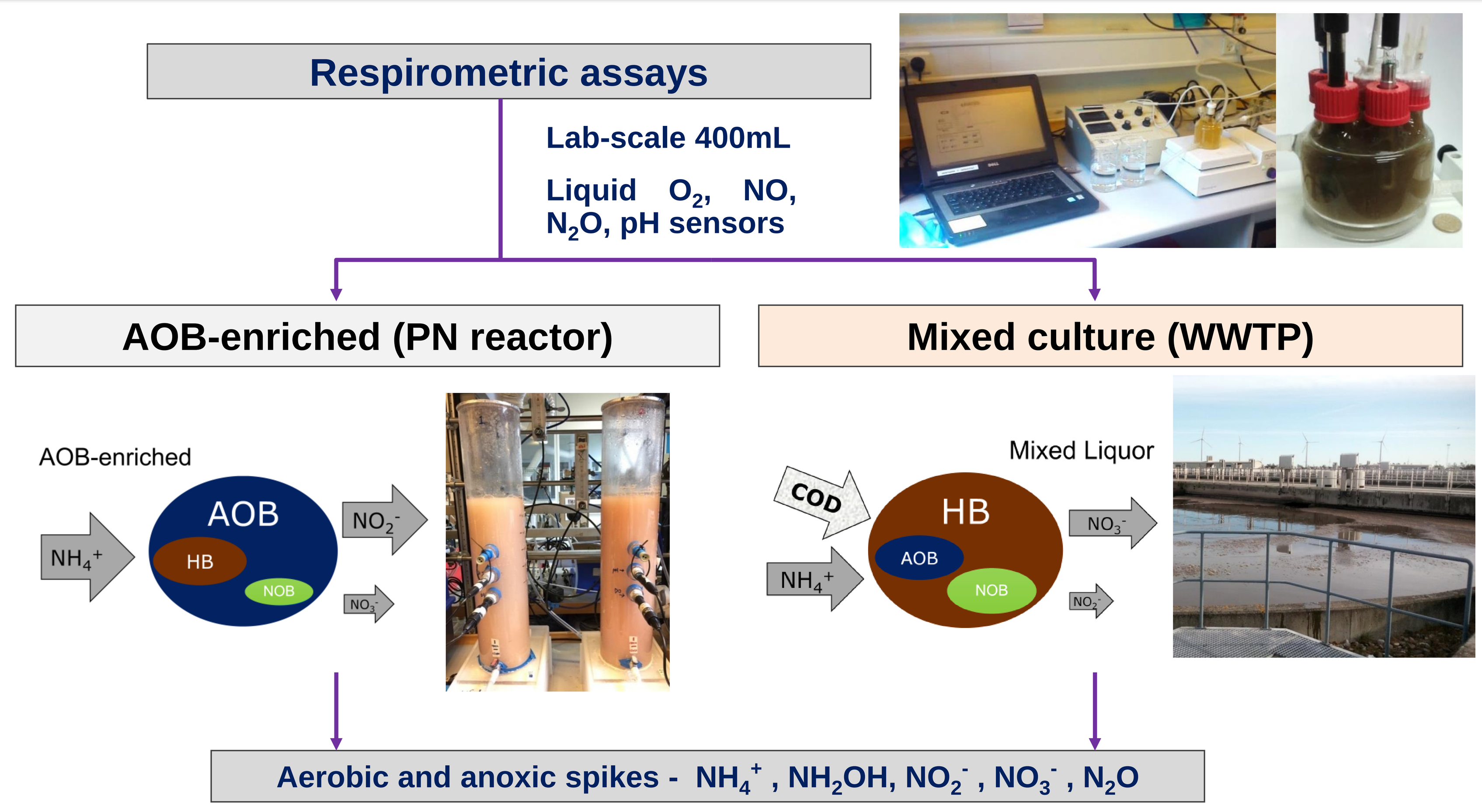
Mixed culture

The mixed culture biomass showed higher specific denitrification rates, N_2O consumption and NOB activity compared to the AOB-enriched.

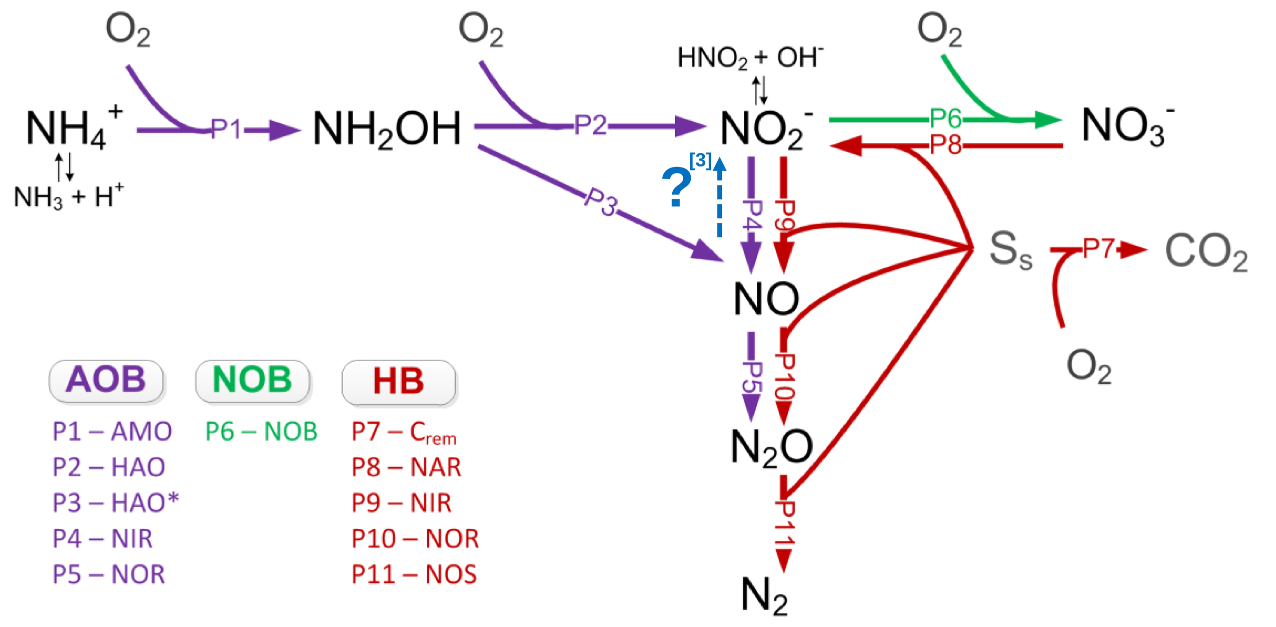


Top – Heterotrophic activity. Denitrification and aerobic carbon removal (left). Maximum N_2O consumption rate at different pH (middle). NO and N_2O dynamics after NO_2^- spike under electron donor limiting conditions (right).
Middle – Aerobic NH_4^+ oxidation. DO consumption (left), NO producion (middle), N_2O production (right).
Bottom – Model evaluation at varying DO and NO_2^- concentrations. From left to right: Pathway contributions to the total N_2O pool NN, ND, HD; N_2O emission factor.

Experimental procedure and Model structure



The NDHA model[1] comprehensively describes N_2O and NO producing pathways by both autotrophic ammonium oxidizing and heterotrophic bacteria:



N2O production pathways

NN - Nitrifier nitrification

ND - Nitrifier denit.

HD - Heterotrophic denit.

Ab - Abiotic*

Other processes

Metabolism of AOB, NOB, HB, Decay, Hydrolysis, Physicochemical, etc.

Model calibration: AOB-enriched vs. Mixed culture

AOB-enriched biomass

DO, NH_4^+, NO_2^-, NO_3^- dynamics

Parameter		Unit	Value
$\mu_{AOB.AMO}$	Maximum AMO-mediated reaction rate	d^{-1}	0.49 ± 0.01
μ_{NOB}	Maximum NOB growth	d^{-1}	0.67 ± 0.07
k_H	Hydrolysis rate	d^{-1}	2.01 ± 0.02
$K_{AOB.NH3}$	NH_3 affinity for AOB	mgN/L	0.12 ± 0.005
$K_{AOB.O2.AMO}$	O_2 AMO-mediated affinity constant	mgO_2/L	0.23 ± 0.02

NO and N_2O dynamics

Parameter		Unit	Value
ε_{AOB}	Reduction factor HAO-mediated reaction rate	(-)	0.48 ± 0.005 ($x10^{-3}$)
η_{NOR}	Reduction factor for NO reduction	(-)	0.16 ± 0.005
$K_{AOB.NH2OH.N}$	NH_2OH affinity for AOB during NO reduction	mgN/L	0.25 ± 0.005
$K_{AOB.HNO2}$	HNO_2 affinity for AOB	µgN/L	0.67 ± 0.03

Mixed culture biomass

DO, NH_4^+, NO_2^-, NO_3^- dynamics

Parameter		Unit	Value
$\mu_{AOB.AMO}$	Maximum AMO-mediated reaction rate	d^{-1}	0.49 ± 0.01
μ_{NOB}	Maximum NOB growth	d^{-1}	1.04 ± 0.05
μ_{HB}	Maximum HB growth rate	d^{-1}	5.15 ± 0.11
$K_{AOB.NH3}$	NH_3 affinity for AOB	mgN/L	0.007 ± 0.0012
$K_{NOB.HNO2}$	HNO_2 affinity for NOB	µgN/L	0.027 ± 0.006

NO and N_2O dynamics

Parameter		Unit	Value
ε_{AOB}	Reduction factor HAO-mediated reaction rate	(-)	0.0031 ± 0.0001
η_{NOR}	Reduction factor for NO reduction	(-)	0.36 ± 0.02
η_{NIR}	Anoxic reduction factor for HNO_2 reduction	(-)	0.22 ± 0.01
$pH_{opt.nosZ}$	Optimum pH for N_2O-reduction	(-)	7.9 ± 0.1
w_{nosZ}	Sinusoidal parameter for N_2O-reduction	(-)	2.2 ± 0.2
$K_{HB.N2O}$	N_2O affinity constant for HB	mgN/L	0.078 ± 0.020

The calibrated model describes N_2O production from AOB-enriched[2] and mixed culture biomass; a total of 10 and 17 parameters were accurately estimated respectively. Parameter sets for each biomass (maximum rates, substrate affinities) highlighted differences in microbial community composition:

- The estimated NH_3 affinity differed, probably due to the different NH_4^+ and pH levels at which the biomasses operated: NH_4^+_AOB-enriched ≫ NH_4^+_Mixed-culture → $K_{AOB.NH3}$ = 0.12 vs. 0.007 mgN/L respectively.
- The fractions of NH_4^+ oxidized (NN pathway) and NO_2^- reduced to N_2O by AOB (ND pathway) also varied between systems → ε_{AOB} = 0.003 vs. 0.0005 (-).

A pH-dependent function to describe N_2O consumption is proposed (max pH = 8).

The NDHA model structure is in agreement with the newly proposed AOB metabolism of aerobic/anoxic NH_2OH oxidation [3].

Conclusions

- A novel experimental design to calibrate N_2O models through extant respirometry is proposed that combines DO, N_2O and NO measurements.
- N_2O and NO production from mixed liquor biomass increased during NH_4^+ oxidation at low DO concentrations and in the presence of NO_2^-.
- The NDHA model response was validated and described N_2O production at varying DO, NH_4^+ and NO_2^- concentrations.
- During NH_4^+ oxidation the NN pathway showed the largest contribution at high DO levels, while the ND and HD pathways increased and dominated the total N_2O production at low DO and high NO_2^- concentrations.

[1] 10.1039/C6EW00179C; [2] arXiv:1705.05962v1; [3] 10.1073/pnas.1704504114

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